UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

NOV 6 1984

Memorandum:

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

SUBJECT: Actual Chlordimeform Residues.

FROM: J. Garbus, Chemist
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THRU: C. L. Trichilo, Chief
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Hazard Evaluation Division (TS-769)

Toxicology Branch is currently reviewing toxicity data for the acaricide-pesticide N'-(4-chloro-o-tolyl)-N,N-dimethylformamide, [chlordimeform, (CDM)]. Two metabolites of this compound, 4-chloro-o-formotoluidide and 4-chloro-o-toluidine, have been found to be oncogenic in rodents. This finding raises the need for quantitative risk assessment for humans.

At present the only use of chlordimeform in the United States is on cotton. Concern arises from residues in cotton seed, in cotton seed by-products and secondary residues as a result of the use of these commodities. Toxicology Branch has requested that the Residue Chemistry Branch provide it with the identification and characterization of all probable residues of chlordimeform and its metabolites in these commodities; an estimation of the real levels of the residues, rather than those established as tolerances, and an estimate of the probable human exposure to each residue. In addition, Toxicology has asked if there are any differences in the residue levels and metabolism of the free base and the salt of chlordimeform. Toxicology also would like guidance in devising a simplified, standardized nomenclature for chlordimeform and its metabolites, preferably one using distinctive acronyms.

Background

Chlordimeform is a member of a novel class of insecticides posessing the formamidine group. It was introduced in 1966 as a broad spectrum acaricide-pesticide active against pests which had acquired resistance to organophosphates and carbamates, but its only current use in the United States is against predators of cotton - the bollworm, the tobacco budworm and the cotton leaf perforator. Chlordimeform is marketed in two forms: as a 48.5% emulsifiable concentrate (EC formulation) and in the form of its hydrochloride salt as a water soluble product (SP formulation).

The current label indicates that chlordimeform is to be used only on cotton, although in the past it was employed on a wide variety of crops for which tolerances were established and never revoked (40 CFR 180.285). In 1976 the product was recalled by its American manufacturers and reintroduced in 1978 with the restriction to its use on cotton at a lower application rate(1 lb. ai/A reduced to 1/4 or 1/8 lb. ai/A).

Tolerances are established for the combined residues of chlordimeform and its metabolites containing the 4-chloro-o-toluidine moiety from the application of the free base or the hydrochloride salt. For the commodities of present interest the tolerances are: cottonseed (5.0 ppm); meat, fat, meat by-products of cattle, goats, hogs, horses, poultry and sheep (0.25 ppm); and milk (0.05 ppm). There is a food additive tolerance for cottonseed hulls of 10 ppm (21CFR 561.80). The usage directions recommend 1/8 to 1/4 lb ai/A on a 3 to 5 day schedule. Application is to continue as long as insect eggs of predators are present. There are no restrictions as to the maximum number of applications per season. The preharvest interval is 21 days.

Nature of the Residue

EC vs SP Formulations

As to one of the concerns raised by Toxicology, all of the available studies indicate no difference in residues resulting from the use of either of the two types of formulation, the free base or the salt. Toxicology infers that greater residues would result from the use of the salt. Its letter states, "CDM is formulated in two forms, a volatile base (CDM-Base) and a relatively non-volatile HCl salt (CDM-Salt). The CDM base applications should result in vaporization dissipation of CDM while the CDM salts should result in higher residues of CDM/metabolites in plant materials."

The description of the base form of chlordimeform as volatile is incorrect, using the commonly accepted meaning of volatile: "readily vaporizable at a relatively low temperature". Chlordimeform base has a M.P. of 35°C, a vapor pressure of 3.5×10^{-4} torr at 20°C and a boiling point of 156°C at a pressure of 0.4 torr. These are the physical characteristics of a relatively non-volatile substance. The decrease in the levels of residual chlordimeform base cannot be attributed to vaporization dissipation but more likely is due to the ready hydrolysis of the compound in neutral or acidic media.

In direct comparisons of chlordimeform salt and base, no differences in residue levels were found. In 1982, NOR-AM submitted residue data from trials on cotton in which the EC and SP formulation were each used in tank-mixes and residue levels on cotton were determined . The most recent summary of chlordimeform residues concludes, "No differences could be shown between the SP and EC formulations...". (C. Blalock, review of residue chemistry for chlordimeform registration standard, 6/8/84).

Metabolites of Chlordimeform

The major metabolites of chlordimeform are demethyl chlordimeform, (also named desmethlyl chlordimeform), 4-chloro-o-formotoluidide, (also known as N-formyl-4-chlorotoluidine), and 4-chloro-o-toluidine. These arise by sequential hydrolytic cleavage of chlordimeform. In addition, other metabolites that have been identified are: didemethyl chlordimeform, 5-chloroanthranillic acid and N-formyl-5-chloroanthranillic acid found in plant and animal tissues: 4-chloro-o-acetotoluidide, (4-chloro-2-methyl-acetanalide); 4-chloro-o-tolylurea; N-methyl 4-chloro-o-tolylurea and N,N-dimethyl 4-chloro-o-tolylurea in animal tissues; 4-chloro-2'methyl malonanillic acid and 4,4'-dichloro-2,2'dimethyl-azobenzene in soil as the result of microbial metabolism of chlordimeform and bis 4-(N,N-dimethyl-N-'o-tolyl formamidine) ether, found in water as the result of photohydrolysis. (See appendix for structural formulas.)

Analytical Methods

The analytical methods that have been used to determine the residues of chlordimeform and its metabolites are based upon the determination of 4-chloro-o-toluidine after prolonged extraction and hydrolysis of plant and animal material. The reported residue levels represent the sum of the levels of chlordimeform and its metabolites that are convertable to 4-chloro-o-toluidine. Metabolites that do not contain this moiety or are resistant to the hydrolytic procedure will not be determined. The current limit of detection is 0.03-0.05 ppm.

Actual Residue Levels

As chlordimeform's only current use is on cotton, only residue data from cottonseed and cottonseed by-products will be discussed. The first question is how much of chlordimeform and its metabolites actually remains as residue in or on cottonseed when chlordimeform is applied according to the registered usage of 1/8 lb to 1/4 lb per acre with an unlimited number of applications per season and a PHI of 21 days.

The data that are available demonstrate that the levels of chlordimeform residues vary considerably with no appparent correlation with amounts applied, geographical areas or PHI's. The only conclusions that are evident are the reasonable ones that residue levels decline with time and that a PHI of 21 days results in residue levels below the established tolerance of 5.0 ppm.

When chlordimeform is used alone, with the current usage pattern, residue levels are about 0.33 ppm with a considerable number of samples reported as having no detectable residues. The highest value reported in any trial was 2.44 ppm. Earlier tests, at different rates of application, using chlordimeform in tank mixes, and employing a less sensitive analytical method resulted in residue levels of about 0.60 ppm with a maximum of 3.1 ppm.

Four cottonseed processing studies contain relevant data. In all of these studies chlordimeform was applied 12 times at a rate of 1/4 lb per acre with a PHI of 21 days. The results given in the following table are averages of the four trials.

Commodity	Residue(ppm)	Range
Cottonseed	0.29	0.20-0.40
Cottonseed Meal	0.21	0.10-0.30
Soapstock	0.21	0.07-0.38
Crude cottonseed oil	0.14	0.07-0.23
Refined cottonseed oil	0.13	0.05-0.22
Refined,bleached, deodorized oil	<0.01	-----
Refined,hydrogenated oil	<0.05	-----
Hulls	0.95	0.50-2.00

Estimates of Actual Residue Levels of Specific Metabolites

The analytical method used to determine these residues of chlordimeform does not determine the level of specific metabolites but only the total level of all compounds convertible to and reacting as 4-chloro-o-toluidine. To arrive at an estimate of the levels of specific metabolite residues, we can use data from radioactive tracer studies of chlodimeform metabolism in cotton. These show that radioactivity from labeled chlordime-

form is found in cottonseed harvested 5 months after the application of the insecticide to the whole plant. Twenty to thirty percent of the radioactivity was recoverable as material reacting as 4-chloro-o-toluidine; 3% as 5-chloro-formyl-anthranilic acid; 5-40% as unextractable from the constituents of cottonseed and the remainder of the recoverable radioactivity as at least 8 unidentified polar compounds. "The conclusion can be drawn from these studies that metabolism in the seed is extensive and continues past the 4-chloro-o-toluidine moiety to hydroxylated compounds that conjugate with natural products and, so far, are not recoverable." (C. Blalock review, 6/8/84) Trials conducted in the early '70's with what are now exaggerated rates (2 lbs/A) resulted in residues high enough to permit the determination of specific metabolites in cotton seed products.

Commodity	Total Residue	Percentage of total residue as:			
	(ppm)	CDM	DeMeCDM	4-Cl-FT	4-Cl-T *
Seed	5.6	-	-	-	-
Hulls	6.53	40	0.5	35	24
Oil(solvent)	0.33	61	9.0	24	6.0
Meal(solvent)	1.48	32	2.0	64	2.0
Oil(screwpress)	1.14	52	2.0	44	2.0

* CDM = Chlordimeform;
4-Cl-FT = 4-chloro-o-formotoluidide;
DeMeCDM = Demethylchlordimeform
4-Cl-T = 4-chloro-o-toluidine

This data shows that about half of the reported residues in cottonseed products is the parent compound and half represents the sum of 4-chloro-o-formotoluidide and 4-chloro-o-toluidine. If we assume that the distribution of metabolites in the by-products reflects the distribution in cottonseed then the reported residues for cottonseed consist of 50% of chlodimeform and 50% of the two substituted toluidines. These, in turn, represent only about 30% of the residual metabolites of chlordimeform; the rest are either unextractable or are not determined by the procedure and remain unidentified.

We can now combine the data and the assumptions to arrive at an estimate of the actual residue levels in cottonseed and cottonseed byproducts for chlordimeform and 4 of its metabolites and for the unextracted, undetermined fraction.

Commodity	Total Residue Found (ppm)	Estimated Residue Level (ppm's)				
		CDM	DeMeCDM	4-Cl-FT + 4-Cl-T	5-Cl-AA	Unextracted Unidentifie
Cottonseed	0.3	0.15	<0.01	0.15	<0.01	0.70
Meal	0.2	0.07	"	0.14	"	0.49
Soapstock	0.2	0.11	"	0.11	"	0.49
Oil, crude	0.15	0.09	0.01	0.04	"	0.04
Oil, R	0.15	0.07	<0.01	0.07	"	0.31
Oil, RBD	<0.01	---	----	----	----	<0.03
Oil, RBDH	<0.05	---	----	----	----	<0.11
Hulls	1	0.38	<0.01	0.57	<0.01	2.22

5-Cl-AA = 5-chloroanthranilic acid
R = refined; B = bleached; D = deodorized; H = hydrogenated

Estimated Actual Residues in Feedstocks

We can use these estimated values together with the data of the composition of livestock feeds given in the residue chemistry guidelines to calculate an estimate of the actual levels of chlordimeform residues that could be presented to animals ingesting feed containing cottonseed, cottonseed meal, soapstock, and hulls. However it is unlikely that livestock diets would contain the maximum possible content of cottonseed and cottonseed by-products. We have also calculated the dietary burden if chlordimeform residues were present at the level of the established tolerance.

Livestock	Potential Level in Feed at Tolerance Level (ppm)	Estimated actual residue in feed (ppm)		
		Reported Residues	Metabolite Residues*	Total Residues**
Beef cattle	3.75	0.26	0.13	0.61
Dairy cattle	2.50	0.16	0.08	0.37
Turkeys and Broilers	1.00	0.07	0.04	0.16
Laying Hens	0.75	0.06	0.03	0.14
Swine	0.75	0.05	0.03	0.12

*Assuming that 4-chloro-o-toluidine and 4-chloro-o-formotoluidide = 50% of the measurable residue. ** Includes unextractable, unidentified components.

Metabolic Disposition of Ingested Residues

Results from studies of administering C^{14} labeled chlordimeform to goats, rats, and dogs indicate that within 96 hrs, 85 to 96% of the label was excreted in urine and feces. Only about 20% of the excreted radioactivity has been identified as known metabolites of chlordimeform. Of these about 0.5% is the unchanged parent compound. Demethyl chlordimeform, 4-chloro-o-formotoluidide, 4-chloro-o-toluidine, and 5-chloroformylanthranillic acid are the major metabolites found in urine.

Very small amounts of radioactivity were found in tissues in these studies. Values from the studies are difficult compare as the amount fed varies as does the time to analysis, the mode of administration, the species, and the expression of results.

Animal	Tissue	% of radioactive dose found in tissue
Goat	Blood	<0.2
	Milk	0.3 - 0.9
	Tissues	NDR
Rat	Blood	0.08 - 0.3
	Kidney	0.1 - 0.3
	Liver	0.03 - 0.5

As with the excreted radioactivity, the bulk of the metabolites in tissues has not been characterized.

Animal feeding studies have been conducted with lactating dairy cattle and with poultry. In these studies animals were fed chlordimeform as part of their ration at the tolerance level. Residues were determined as the 4-chloro-o-toluidine moiety. No residues were found in milk, muscle or eggs. Residues were detected in liver, kidney, and chicken fat at levels equivalent to 0.1 to 0.2% of the daily dose.

We can use these figures to estimate the potential actual residues and secondary residues of chlordimeform in items of the diet for which there are data. It should be realized that these are estimates derived by calculation and inference from limited and often unreplicated data, that there is large variability in the reported residue data, that usage patterns have been changed, and that analytical methods differ. Also, the residue methodology has a limited sensitivity (0.03-0.05 ppm); the methodology determines only about 30% of the metabolites of chlordimeform; and the amount and nature of the undetermined residue remains unknown.

Dietary Item	Chlordimeform Metabolite Residues	
	micrograms/Kg	ppm
Cottonseed Oil, Refined	130	0.13
" " RBD	<10	<0.01
" " RBDH	<50	<0.05
Beef Liver	2.4	0.002
Chicken Liver	4.6	0.005
Chicken Fat	24.7	0.025
Goat's Milk	6.7	0.007

For other items of dietary interest, eggs, cow's milk, beef and chicken muscle, there are no reported secondary residue levels as these were below the detectable limits of the analytical methods. Therefore estimates of actual residues of chlordimeform in these items cannot be made. However, we assume that they will be in the same range as those estimated for the dietary items in the table.

We also assume that half of the estimated residues will consist of 4-chloro-o-formotoluidide plus 4-chloro-o-toluidine.

Note on Nomenclature

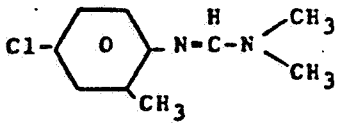
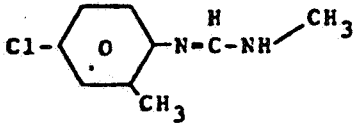
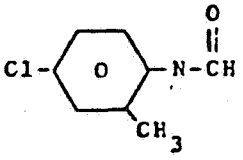
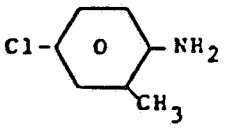
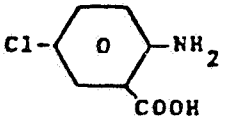
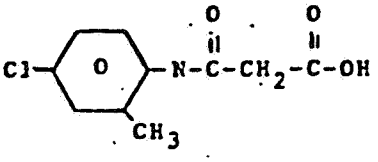
We prefer that the systematic or common usage name of a chemical be used. This nomeclature is standardized, is descriptive and aids in understanding and following chemical reactions and metabolic transformation. Abbreviations and acronyms can be idiosyncratic and ambiguous and often cannot be readily translated to other terms used for the same compound. However if abreviations are desired, they should be informative and descriptive in themselves. To that end, we have included an appendix showing the structures of chlordimeform and its metabolites, their systematic names and suggested abbreviations.

cc: S.F., R.F., Circ., G. Beusch, Reviewer, PM-12
RDI:A.A. Rathman:10/31/84:R. D. Schmitt:10/31/84
TS-769:RCB:CM#2RM810:X7484:J. Garbus:edited:wh:11/2/84

APPENDIX

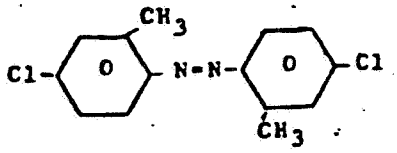
CHLORDIMEFORM

Identified Metabolites and Degradation Products

Nomenclature	Suggested Abbreviations
 chlordimeform	CDM
 desmethylchlordimeform	DeMeCDM
 4-chloro-o-formotoluidide	4-Cl-o-FT
 4-chloro-o-toluidine	4-Cl-o-T
 5-chloroanthranilic acid	5-Cl-AA
 4'-chloro-2'-methyl-malonanilic acid	4-Cl-Me-MaA

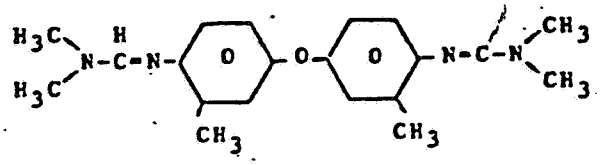
Nomenclature

Suggested Abbreviations



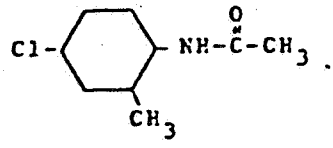
4,4'-dichloro-2,2'-dimethylazobenzene

DiClDiMeAz



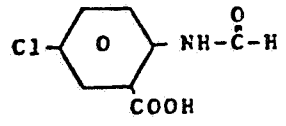
bis-4-(N,N-dimethyl-N'-o-tolyl formamidine) ether

DiMeoTFE



4-chloro-o-acetotoluidide

4-Cl-o-AcT



5-chloro-formylanthranillic acid

5-Cl-FAA